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Population studies of an endemic gastropod from waterfall environments

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Abstract: Chilinidae is a family endemic to South America, ranging from the Tropic of Capricorn to Cape Horn and the Falkland Islands, and includes 32 species. However, there are few population studies on the Chilinidae. We study aspects of the ecology of an endemic species, *Chilina megastoma* Hylton Scott, 1958, from the Arrechea Falls in the Iguazú National Park, Argentina, such as density and individual annual growth trends. Nine samplings were carried out between December 2003 and December 2005, using two transects that crossed the waterfall. Individual annual growth rate was analyzed according to length, following von Bertalanffy´s model. Six cohorts were identified, some in the same climatic season but successive years (two in winter and two in summer). The winter and autumn cohorts reached 85% of their last whorl length in the first year. Compared to other families of gastropods from subtropical climates, these populations have several recruitment events per year, but never in winter.

Key words: Argentina, *Chilina*, growth rate, Iguazú Falls, von Bertalanffy model

In Argentina, there are forty (40) endemic species for freshwater Gastropoda: Thiaridae (3 species), Ampullariidae (1), Cochliopidae (10), Lithoglyphidae (11), Chilinidae (11), Lymnaeidae (2), and Physidae (2). Of the species of Lithoglyphidae, Cochliopidae, and Chilinidae, 50, 62.5, and 68.7 %, respectively, are endemic (Rumi *et al.* 2006). Although most of these endemic species are considered vulnerable to extinction, they are in serious need of bio-ecologic studies to allow more precision in the identification of their conservation status.

Chilinidae (Pulmonata, Basommatophora) is a family exclusive to South America, ranging from the Tropic of Capricorn to Cape Horn and the Falkland islands. The family includes a single genus, *Chilina* Gray, 1828 with about 32 species, 17 of which have been recorded in Argentina (Castellanos and Miquel 1980, Castellanos and Gaillard 1981, Gutiérrez Gregoric and Rumi 2008) while the rest are distributed in Chile.

Many Argentinean species of *Chilina* are endemic, and their biology and ecological strategies are practically unknown. In general, only a few studies have been recently reported in this family: Miquel (1986) studied the life cycle of *Chilina fluminea* (Maton, 1809) and its gonad evolution; Bosnia *et al.* (1990), analyzed the growth of *Chilina gibbosa* G. B. Sowerby I, 1841, and its density; Estebenet *et al.* (2002) analyzed the natural diet for *Chilina parchappii* (d'Orbigny, 1835). Quijon and Jaramillo (1999) and Quijon *et al.* (2001), worked on *Chilina ovalis* (Sowerby, 1841) from southern Chile, emphasizing spatial distribution and growth.

The dominant landscape in Iguazú National Park (INP) includes waterfalls and rapids of the Iguazú River, all surrounded by subtropical forest. The park is home to four endemic species of molluscs: *Eupera iguaznensis* Ituarte, 1989 and *Eupera elliptica* Ituarte and Dreher-Mansur, 1993 (Bivalvia: Sphaeriidae), and *Chilina megastoma* Hylton Scott, 1958 and *Chilina iguaznensis* Gutiérrez Gregoric and Rumi, 2008 (Gastropoda: Chilinidae). *Chilina megastoma* and *Acrorbis petricola* Odhner, 1937 (Planorbidae) are exclusively found on waterfalls. *Acrorbis petricola* has been recorded at only three localities, one in Brazil and the other two in Argentina—one in grounds of the INP. Upstream dam and reservoir construction during recent years have modified the hydrologic regime of the river. These changes could induce irretrievable losses in biodiversity, especially among these types of environments. In this sense, a paradigmatic example is that of *Aylacostoma* Spix, 1827 (Gastropoda: Thiaridae), which included four described species known only in the rapids along the upper Paraná River, in the sector now occupied by the Yacyretá Reservoir (Argentina – Paraguay). According to Quintana and Mercado Laczkó (1997) these species can be considered extinct in their natural habitat. The same may have occured with *Chilina gnaraniana* Castellanos and Miquel, 1980 and *Acrorbis* sp., also collected before the flooding of the reservoir (Rumi 1986, Rumi *et al.* 2006).

The scarce information available on gastropods from white-water rivers with waterfalls such as the Iguazú deals with those that lived upstream and downstream from waterfalls, or with the description of new species from this kind of environment (Ponder 1982, Glöer *et al.* 2007). However, population dynamics of these species remain largely unknown.

This work focuses on estimating population patterns such as density, individual growth rate, and recruitment times of a population of *Chilina megastoma*. This species is endemic to

the waterfalls in the Iguazú National Park, Misiones province, Argentina.

MATERIALS AND METHODS

Study area

Iguazú National Park is located in Misiones province, at the northeastern corner of the country. This park was chartered in 1934, and in 1984 was declared a World Natural Heritage Park because of the high biological diversity of the subtropical Paranensean Forest and the numerous waterfalls that average 75 m high. The climatic characteristics of INP (1600 mm annual precipitation, 21.1 °C mean annual temperature) allow wide habitat diversity and support a varied flora and fauna. The waterfalls harbor plants and animals that are especially adapted to the constant humid conditions and the force of water. Abundant islands covered by a distinct type of forest exist along the upper course of the Iguazú River. The population study was done in the Arrechea Falls of the INP (25°39'S, 54°27'W) (Fig. 1).

Sampling methodology

The study comprised nine seasonal samplings between December 2003 and December 2005 (December 2003, 2004, 2005; February 2004, 2005; June 2004, 2005; September 2004, 2005). Water temperature (°C), conductivity (µS), hardness (°f, calculated as conductivity / 20), total dissolved solids (mg/l), pH, dissolved oxygen (mg/l), and saturated oxygen (%) were measured during most samples.

Two transects, each approx. 10 meters long, were followed from the bottom of Arrechea Falls behind the waterfall, and into the driest vegetated zone at the top (Fig. 1). The snails were hand collected from rocks exposed along the two transects. Squares of 0.15 m (0.0225 m^2), placed at 0.40 m along the transect, were employed as a sample unit (SU). For each sampling date, we calculated the average density of snails and its standard deviation.

Analysis of growth

For individual growth rate, snails were measured on site using 0.01 mm precision calipers. As the apex of these gastropods is usually damaged by water current, only the length of last whorl (LWL) was recorded. Once measured, snails were returned to their natural environment. In all samples, size frequency intervals of 1 mm were used. The size frequency distribution of each sample was separated by its modes, which were assumed to correspond to coexisting cohorts. Frequency distributions corresponding to each cohort were fitted to a normal curve, for which mean and standard deviation were calculated. These values were used in the growth analysis. Individual growth rate was analyzed according to length,

Figure 1. Transects in the Arrechea Falls, Iguazú National Park, Argentina.

Table 1. Mean water quality parameters at Arrechea Falls. Cond, conductivity (µS); TDS, total dissolved solids (mg/l); T, temperature (°C); DO, dissolved oxygen (mg/l); Sat, % oxygen saturation; n/d, no data.

Date	Cond	TDS	T	pH	DO	Sat
Dec-03	490	25.8	23.3	6.5	4.9	69.3
Feb-04	95.2	42.6	22.4	6.6	6.5	73
Jun-04	235.4	122.6	14.5	7.3	8.7	81
Sep-04	57.2	29	20.4	7	9.2	101
Dec-04	39.7	19.8	21.3	6.8	8.4	94
Feb-05	137.9	65.7	23.7	8.3	5.3	62.5
Jun-05	66	32.2	18.6	6.6	n/d	n/d
Sep-05	n/d	n/d	14.5	7.4	2.4	24
Dec-05	n/d	n/d	n/d	n/d	n/d	n/d

Table 2. Mollusc density (δ: ind/m^2), including *Chilina megastoma* oviposition density (ovipositions/m^2). *N*, total individual; $\bar{x}$, mean calculated per SU (sample unit = 0.0225 m^2); *SD*, standard deviation.

Date	*N* of SU		*Chilina megastoma*	*C. megastoma* ovipositions	*Acrorbis petricola*	*Potamolithus* spp.	*Succinea* sp.	*Uncancylus concentricus*
Dec-03	45	*N*	233	14	787	75	9	0
		$\bar{x}$	5.18	0.31	17.49	1.67	0.20	0
		SD	5.39	0.84	13.75	4.7	0.68	0
		δ	230	14	777	74	9	0
Feb-04	51	*N*	184	21	829	121	17	0
		$\bar{x}$	3.61	0.41	16.25	2.37	0.33	0
		SD	4.17	1.11	20.55	5.78	1.07	0
		δ	160	18	722	105	15	0
Jun-04	50	*N*	210	24	296	378	4	2
		$\bar{x}$	4.2	0.48	5.92	7.56	0.08	0.04
		SD	6.86	1.74	7.31	23.01	0.27	0.4
		δ	187	21	263	336	4	2
Sep-04	53	*N*	176	11	629	244	45	3
		$\bar{x}$	3.32	0.21	11.87	4.6	0.84	0.06
		SD	5.25	0.79	17.62	13.5	2.04	0.42
		δ	148	9	527	205	38	2
Dec-04	50	*N*	251	9	632	256	5	0
		$\bar{x}$	5.02	0.18	12.64	5.12	0.1	0
		SD	7.5	0.6	24.06	11.53	0.46	0
		δ	223	8	562	228	4	0
Feb-05	26	*N*	53	6	142	556	1	0
		$\bar{x}$	2.04	0.23	5.46	21.38	0.04	0
		SD	2.44	0.65	7.66	21.94	0.2	0
		δ	91	10	243	950	2	0
Jun-05	51	*N*	35	1	254	467	2	0
		$\bar{x}$	0.69	0.02	4.98	9.16	0.04	0
		SD	1.47	0.14	12	23.82	0.2	0
		δ	30	1	221	407	2	0
Sep-05	45	*N*	74	15	338	124	55	0
		$\bar{x}$	1.64	0.33	7.51	2.75	1.22	0
		SD	6.84	1.65	22.11	8.38	2.8	0
		δ	73	15	333	122	54	0
Dec-05	43	*N*	79	10	113	120	16	2
		$\bar{x}$	1.84	0.23	2.63	2.79	0.37	0.05
		SD	4.25	1.38	4.49	5.59	1.07	0.3
		δ	82	10	117	124	17	2

following von Bertalanffy's model (1938). This model has been widely applied for studies on Planorbidae (Gastropoda, Pulmonata) populations either for experimental designs on site, in the laboratory, or under natural conditions (Loreau and Baluku 1987, Baluku and Loreau 1989, Ituarte 1989, 1994, Rumi *et al.* 2007).

The model is:

$$LWLt = LWL\infty\ (1 - e^{-k\ (t-t_0)}), \quad (1)$$

where $LWL\infty$ = maximum length of last whorl, k = growth rate constant, t = time, and t_0 = hypothetical time in which length = 0. From linear transformation of the logarithmic equation (1) we obtained:

$$ln\ (1\text{-}\ LWLt/LWL\infty) = -kt + kt_0 \quad (2)$$

A regression equation between $ln\ (1\text{-}\ LWLt/LWL\infty)$ and t was calculated. In (2), kt_0 = origin of ordinate (a), $t_0 = a/k$, and the slope (b) = (–)k.

Time measured for each sample was divided into parts of one year, following Basso and Kher (1991) and Rumi *et al.* (2007), in the equation:

$$T = [(month - 1)*30 + sampling\ day] / 360 + A,$$

where A = sampling year. The year in which the study started was considered as $A = 0$, the following year is $A = 1$, etc. Thus, $t = 0$ corresponded to January 1st, $t = 0.5$ to July 1st, and $t = 1$ corresponds approx. to December 31st. Maximum length of the whorl was calculated on mean values of cohorts obtained from decomposition, using Walford's method (1946).

Only means from cohorts beginning at the time of sampling were considered; the cohorts with the two highest values (December 2003 and February 2004) and lowest values (June, September, and December 2005) were not considered. In order to compare these data with those from other species or from the same species in different environments, t_0 was considered as 0 and growth rate was expressed as percentage of maximum length of last whorl, as reported by Rumi *et al.* (2007).

RESULTS

The averages of water quality parameters were: temperature ($N = 8$) = 19.8 °C ($SD = 3.7$); pH ($N = 8$) = 7.1 ($SD = 0.6$); conductivity ($N = 7$) = 160.2 µS/cm ($SD = 159.8$); total dissolved solids (TDS) ($N = 7$) = 48.2 mg/l ($SD = 36.7$); dissolved oxygen ($N = 7$) = 6.5 mg/l ($SD = 2.5$); oxygen saturation = 72.1% (Table 1). Water hardness (°f) was 8.01 (soft water).

Gastropod assemblages in addition to *Chilina megastoma* in the Arrechea Falls included: *Potamolithus* sp. 1 and *Potamolithus* sp. 2 (Lithoglyphidae); *Uncancylus concentricus* (d'Orbigny, 1835) (Ancylidae); *Acrorbis petricola* (Planorbidae); and *Succinea* sp. (Succineidae). *Acrorbis petricola* had the greatest densities, whereas there were only isolated specimens of *U. concentricus* in three samples (Table 2). Individuals of *C. megastoma* from classes 2 to 4 (1 to 3 mm) were observed throughout the entire year, except for February and June of 2005 (Fig. 2). Specimens from classes 10 to 14 (9 to 13 mm) were also observed year round at similar frequencies (Fig. 2).

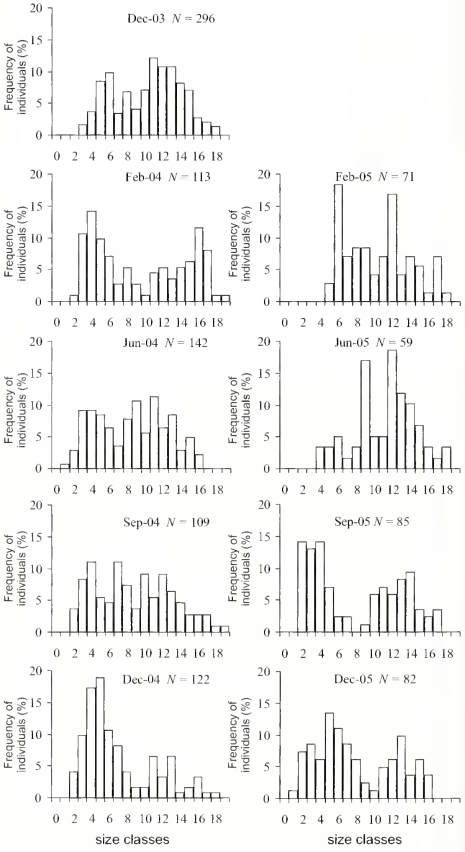

Figure 2. Size-frequency distributions of *Chilina megastoma*, expressed as a percentage of the sample total N, in intervals of 1 mm, among samples at Arrechea Falls.

Table 3. Means ($\bar{x}$), standard deviation (*SD*), and number of individuals (*N*) fitted to each monthly shell-size frequency distribution for cohorts of *Chilina megastoma* in Arrechea Falls.

	Cohort I			Cohort II			Cohort III			Cohort IV			Cohort V			Cohort VI		
Date	$\bar{x}$	*SD*	*N*	$\bar{x}$	*SD*	*N*	$\bar{x}$	*SD*	*N*	$\bar{x}$	*SD*	*N*	$\bar{x}$	*SD*	*N*	$\bar{x}$	*SD*	*N*
Dec-03	7.73	0.62	34	4.79	0.93	76												
Feb-04	12.21	1.07	20	7.31	0.74	12	3.76	1.06	48									
Jun-04	14.54	0.76	6	11.32	1.04	50	7.9	1.08	39	3.49	1.15	49						
Sep-04	16.02	1.35	11				11.21	1.4	40	6.95	0.81	25	3.37	1.08	33			
Dec-04				15.2	1.13	9				11.23	1.36	23	6.39	0.66	21			
Feb-05							16.21	0.90	7				12.04	1.36	31	5.6	0.5	20
Jun-05										16.27	1.08	6				8.65	0.8	16
Sep-05													15.95	0.72	6	11.79	1.6	34
Dec-05																14.77	0.7	9

Polymodal size frequency distributions indicated three or four well-defined cohorts (Table 3 and Fig. 3). Six cohorts were identified (Table 4). The $LWL\infty$ calculated was 18.47mm. Cohorts I and V correspond to winter recruitment, and cohorts III and VI correspond to summer recruitment. Considering the information of both seasonal cohorts together, the growth equations for these two climatic seasons (summer and winter) are given in Table 5.

In all cases the slope (*b*) of the linear regression comparing the observed and calculated data was near 1, and the same was true for R^2: winter cohort b: 1.01, R^2: 0.96; spring cohort b: 0.97, R^2: 0.99; summer cohort b: 1.01, R^2: 0.95; and autumn cohort b: 1.03, R^2: 0.99; indicating a good adjustment of data to the pattern of calculated growth.

The maximum *LWL* growth percentage indicates that all the cohorts reach at least 76% of their growth in the first year of life (Fig. 4). The winter and autumn cohorts reach 85% of their *LWL* in the first year. After two years, all the cohorts reach 94% of their *LWL*, with the winter and autumn cohorts reaching the highest percentage in the two years (97%).

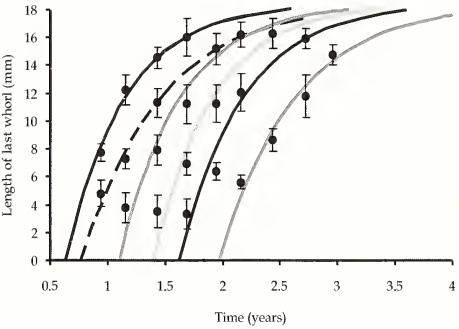

Figure 3. Means (dots) and standard deviation (bars) of the normal curves fitted to each monthly shell-size frequency distribution, and growth curves for *Chilina megastoma* in Arrechea Falls. Lines represent theoretical growth curves according to von Bertalanffy models: black lines represent winter cohorts; dark gray represents summer cohorts; light gray represents autumn cohort; dashed line represents spring cohort.

DISCUSSION

Each of the molluscs has a specific distribution pattern. *Potamolithus* spp. inhabit mainly the pool and to a lesser extent the water trapped among the rocks. *Succinea* sp. inhabits higher places where rocks begin to be colonized by vegetation. *Acrorbis petricola* and *Chilina megastoma* may be found along the whole height of the waterfall; the former lives on the vertical rocks of the wall, while *C. megastoma* dwells generally in the crevices and in areas more strongly hit by water.

The decrease in density of *Chilina megastoma* and *Acrorbis petricola* beginning in February of 2005 was due to the drought recorded in the Iguazú area from December 2004 to May 2005 (Center of Subtropical Ecological Investigations–CIES, unpubl. data) (Table 2). The two species inhabit rocky and humid sectors of the waterfall, which, because of the low water level of the river, remained dry. In samples in February and June 2005 (those most affected by drought) there were no specimens of the smaller size classes. There may have been insufficient humidity to allow oviposition or keep hatchlings viable.

Table 4. Exponents for the growth equations for each cohort. k, growth rate constant; t_0, hypothetical time in which length = 0.

Cohort	k	t_0
Cohort I	1.93	0.63
Cohort II	1.46	0.77
Cohort III	1.88	1.10
Cohort IV	1.96	1.42
Cohort V	1.82	1.62
Cohort VI	1.52	1.97

Table 5. Exponents for the growth equations for each seasonal cohort. k, growth rate constant; t_0, hypothetical time in which length = 0.

Seasonal cohort	k	t_0
winter	1.88	0.63
spring	1.46	0.77
summer	1.73	0.05
autumn	1.96	0.40

Populations of *Potamolithus* were not affected by the drought as they were mainly in the pools, which did not dry. During the last two samples (September and December 2005), the population of *C. megastoma* began recovering with an increase in density, and a predominance of juvenile specimens.

Hydrology of the Iguazú River and its tributaries are not only affected by rainfall but also are regulated by hydroelectric projects located upstream, as mentioned above. *Chilina megastoma* turned out to be the most vulnerable species because of its high humidity requirements, because the species does not migrate towards the water pool zone, and because of the long hatching period (25 days; pers. obs.).

Oviposition by *Chilina megastoma* occurred throughout the year, and averaged between 11 and 14.5 per square meter. *Chilina megastoma* may reproduce continually because of the low temperature variation along the year in the Iguazú National Park. Species of Chilinidae in cold areas reproduce once a year. *Chilina gibbosa*, in the Ramos Mexía reservoir (provinces of Río Negro and Neuquén, Argentina) and in Lake Pellegrini (Río Negro province), reproduces during the summer (Bosnia *et al.* 1990, Gutiérrez Gregoric *et al.* 2004, respectively). *Chilina fluminea*, from the "La Balandra" beach, Berisso (Buenos Aires province, Argentina), reproduces only in winter (Gutiérrez Gregoric 2008). *Chilina ovalis*, in southern Chile, does so in spring (October) (Quijon and Jaramillo 1999, Quijon *et al.* 2001). For *C. ovalis* and *C. gibbosa* from Lake Pellegrini, the estimated lifespan for each species is about three years. For *C. fluminea*, Gutiérrez Gregoric (2008) estimated a lifespan of between two and two and a half years, while in *C. megastoma*, longevity is approx. two years (Fig. 4). These two different trends can be due to factors such as temperature. While INP shows little variations throughout the year, in the other environments the seasons are very marked. For *C. megastoma*, its best reproductive period is when temperatures are around 15 °C (Table 1), similar to *C. fluminea* in "La Balandra" beach, where temperatures are near 12 °C.

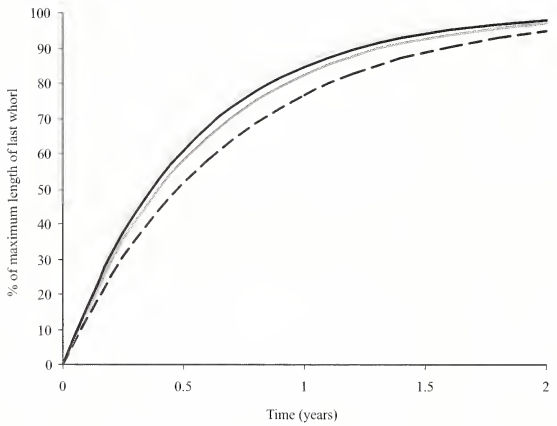

Figure 4. Growth of *Chilina megastoma* expressed as percentage of maximum length of the last whorl.

The growth constants for *Chilina ovalis* and *C. gibbosa* were similar and lower than those of *C. fluminea* and *C. megastoma*. This can be explained by the temperature drop recorded in winter (10 °C for *C. ovalis* in Chile) and the greater longevity in these species.

Other families of gastropods from subtropical climates can have several recruitments per year, but never in winter (Ituarte 1989, 1994, Rumi *et al.* 2007). In *Biomphalaria occidentalis* Paraense, 1981 (Planorbidae) (Corrientes province, Argentina), the growth percentage of the first year of life was always greater than 80%. *Biomphalaria straminea* (Dunker, 1848) and *Biomphalaria tenagophila* (d'Orbigny, 1835) from Artigas, Uruguay reached 80% of maximum length at nine and a half and eight months respectively (Ituarte 1989, 1994). Chilinidae in subtropical environments thus possess slower growth rates than

Planorbidae, but reproduces continually, while the Planorbidae cease reproduction in winter.

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